Cannabinoids as a Promising Therapeutic Approach for the Treatment of Glioblastoma Multiforme: A Literature Review

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ABSTRACT

Glioblastoma multiforme (GBM) or grade 4 astrocytoma is the most malignant form of primary brain tumor. Treatment of glioblastoma is difficult despite of surgery, radiotherapy and chemotherapy. Patients with glioblastoma survive for less than 12 months. Considering to biology function of glioblastoma, researchers have recently offered new therapeutic approaches such as cannabinoid therapy for glioblastoma. Cannabinoids are active compounds of Cannabis sativa that operate in the body similar to endogenous canabinoids –the endocannabinoids- through cell surface receptors. It is interesting that cannabinoids could exert a wide spectrum from antiproliferative effects in condition of the cell culture, animal models of glioblastoma and clinical trials. As a result, Cannabinoids seem to modulate intracellular signaling pathways and the endoplasmic reticulum stress response in glioma cells. Those play antitumoral effects through apoptosis induction and inhibition of glioblastoma angiogenesis. The goal of this study was to discuss cannabinoid therapy and also what cellular mechanisms are involved in the tumoricidal effect of the cannabinoids. In this review article, we will focus on cannabinoids, their receptor dependent functional roles against glioblastoma acccording to growth, angiogenesis, metastasis, and future purposes in exploring new possible therapeutic opportunities.

Keywords: Cannabinoids; Glioblastoma multiforme; Apoptosis; Angiogenesis inhibitors; Clinical trial

INTRODUCTION

Glioblastoma multiforme (GBM), or grade IV astrocytoma, is the most malignant and aggressive forms of primary brain tumors. Cellular and molecular heterogeneity of GBM is a hallmark property for its malignancy and recurrency. Prevalence of glioblastoma was reported 65 percent of astrocytic tumors and it has been estimated as 3-4 cases per 100000 population per years worldwide [1-3]. Results indicated that adults involved in GBM more than young and also higher incidence in men than women [3]. The histopathology characterizations of glioblastoma are nuclear atypia, mitotic activity, cellular pleomorphism, vascular thrombosis, microvascular proliferation and necrosis [3]. Diagnosis and treatment of glioblastoma was failed owing to properties of diffuse infiltration and intratumoral heterogeneity [4]. Glioblastoma complex structure including cancer stem cell subpopulation and genetic lesions has shared in weak diagnosis and treatment [1,5]. Metastasis as a recurrence factor was mediated through the transforming growth factor-beta (TGF-β) and Akt signaling pathways in glioma cells [6,7]. Also, a thick extracellular matrix was produced in glioblastoma invasion to cells migration and proteolytic enzymes digest matrix for enhancing

invasion [3]. Glioblastoma has high proliferation and invasiveness rate which is thought to be the main reasons for resistance to current therapeutic strategies including surgery, chemotherapy, radiotherapy and combination of surgical resection-radiotherapy. Survival rate of patients with glioblastoma is normally 6-12 months after diagnosis and treatment but with high recurrence. Glioblastoma recurrence can occur in patient's brain owing to rapid metastasis of tumor to the neighboring brain structures thereby escaping surgical resection [1]. It is interesting that glioblastoma metastasis is very rare to outside of central nervous system because existence of the subarachnoidal space and cerebrospinal fluid (CSF) [3].

Recently, researchers are surveying new methods such as gene therapy and immunotherapy as safe and new approaches for inducing apoptosis of glioma cells although clinical results have been not succeed yet [8,9]. According to expression of cannabinoid receptors in glioblastoma, numerous studies were demonstrated that cannabinoids through its receptors act as tumoricidal agents for the treatment of glioblastoma [10-12]. In present study, we aim to review the different properties of cannabinoids as therapeutic agents for the management of glioblastoma. A preliminary list of articles was obtained through PubMed and scopus databases. Our search keywords were glioblastoma multiforme, cannabinoid and clinical trial. According to keywords, papers in the English language were considered for review. Finally, papers were studied and interpreted considering to our aim.

LITERATURE REVIEW
The endocannabinoid system

Cannabis sativa has over 400 chemical elements containing alkaloid derivatives of spermidine, sterols, terpens and flavonoid glucosides that have either psychoactive or non psychoactive traits. Δ^9-tetrahydrocannabinol (THC) is the most important chemical compound of cannabinoid [13]. THC plays various biological effects like anandamide and 2-arachidonoylglycerol –the endocannabinoids- via specific cannabinoid receptors [14]. There are two types of cannabinoid specific receptors, CB1 and CB2, in mammalian tissues [15]. CB1 cannabinoid receptor is abundant in specific areas of the brain neurons including nerve terminals for endocannabinoid mediated neuromodulation [16]. In contrast, CB2 cannabinoid receptor was initially distinguished in cells of immune system. However, recent data have been reported the presence of CB2 cannabinoid receptor in brain cells including microglial cells, astrocytes, some neurons, and

glioma cells [17]. Pharmacological studies have indicated that cannabinoid receptor function is the inhibition of adenylyl cyclase in glioma cells. In addition to, CB1 cannabinoid receptor can modulate ion channels via inhibition voltage-sensitive calcium (Ca^{2+}) channels and activation of inwardly rectifying potassium (K^+) channels [15,18]. Cannabinoid receptors were also involved in several signaling pathways for example c-Jun N-terminal kinase and p38 mitogen-activated protein kinase [19,20], extracellular signal-regulated kinase (ERK) [21], focal adhesion kinase (FAK) [22], phosphatidylinositol 3-kinase (PI3K)/Akt [23], and the sphingomyelin cycle [24] in order to control of cell proliferation and survival.

Tumoricidal Activity of Cannabinoids

Several clinical trials have been known that cannabinoids exert alleviative effects including nausea and emesis in cancer patients exposed to chemotherapy. In addition, cannabinoids are testing appetite stimulation and analgesia properties in cancer patients [25,26]. It has further been suggested that cannabinoids may act as tumoricidal agents according to experiments of cell culture and animal models of brain tumor. At first, Guzman has reported the tumoricidal effect of THC in glioma cells via break down of cellular sphingomyelin for the purpose of ceramide generation for inducing apoptosis [27]. Also, animal models demonstrated that local administration of THC lead to decrease of brain tumor size and increase survival rate in rats [28]. Molecular studies on glioblastoma biopsies have been indicated expression enhancement of cannabinoid receptors –CB1 and CB2- in glioma cells. cannabinoid agonists can modulate key signaling pathways including induction of apoptosis in glioma cells through receptor. In addition to, those may inhibit angiogenesis and metastasis in glioblastoma. So, it seems to administrate of selective agonists of cannabinoid receptor could be affected for patients with glioblastoma [10,11].

Induction of apoptosis

Cannabinoids through its receptors can trigger apoptotic signaling pathways in glioma cells [28,29]. Different molecular mechanisms are involved in the apoptosis induction of glioma cells such as production of the pro-apoptotic sphingolipid ceramide [28,30], upregulation of stress-regulated protein via transcription factor of p8, functional alteration of endoplasmic reticulum (ER) and caspase-3 activation via mitochondrial dysfunction. Studies have indicated that ceramide levels have inverse correlation with malignant grade of human glioma tumors [31]. Ceramide synthesis stimulates the expression

of p8 -a transcription factor- in glioma cells. Expression of p8 lead to up-regulation of two transcription factors of ATF4 (Activating Transcription Factor 4) and CHOP (C/EBP homologous protein) in endoplasmic reticulum stress response in order to expression increase of the stress-regulated pseudo-kinase TRB3 (tribbles 3) [32-35]. Convergence of pro-apoptotic protein of ceramide and endoplasmic reticulum stress response are parallel with apoptotic pathway of mitochondria including decrease of mitochondrial membrane potential and the activation of caspase-3 in glioma cells [36-39]. Researchers have focused on relation of the cannabinoid-ceramide for therapeutic applications of glioblastoma. Question proposed which cannabinoid receptor type shared in the apoptosis and antiproliferative effects of glioma cells. In some studies suggested that cannabinoids take effect via two receptors of type 1 and 2 [40] and in other researches either CB1 receptor or CB2 receptor is effective [41,42]. However, expression level of receptor and receptor type were related to apoptosis induction of glioma cells [43]. According to expression of cannabinoid receptors in glioma cells, it is possible to share the endogenous cannabinoids, Anandamide (AEA) and 2-Arachidonoylglycerol (2-AG), of brain for inducing apoptosis in glioma cells [44].

Inhibition of tumor angiogenesis using cannabinoids

Angiogenesis is an important event in growth and metastasis of malignant brain tumors such as glioblastoma. New vascular was repeatedly formed in tumor mass in order to cell nutrition and gas exchange. So, angiogenesis inhibition could be considered as an effective therapeutic approach for glioblastoma. It is essential that we know various cellular and molecular functions of the angiogenesis in glioblastoma including extracellular matrix damage, proliferation of endothelial cells and morphological differentiation of endothelial cells. All these actions were controlled by stimulatory and inhibitory signals such as integrins, angiopoietins, chemokines, growth factors and etc. Immunohistochemical studies in animal models of glioma have indicated that cannabinoids can convert blood vessels of tumor to small and impermeable capillaries [45]. Also, cannabinoids administration can decline expression of vascular endothelial growth factor (VEGF) and its receptor and proangiogenic cytokines such as angiopoietin-2, suggesting that these changes lead to decrease of tumor size [46,47,48]. Pharmacological blockade of cannabinoids in tumors was performed via ceramide production in glial cells of tumor. Ceramide plays the antitumoral and

antiangigenic roles and reduces VEGF expression in vitro and in vivo [47], indicating that ceramid is important in tumoricidal action. Other functions of cannabinoids are related to the inhibition of invasiveness, migration of vascular endothelial cells and apoptosis induction in the endothelial cells [49]. Moreover, cannabinoid using decline the expression of matrix metalloproteinase-2 (MMP2) – a proteolytic enzyme- in mice models of glioma [45,50]. Clinical trial studies have demonstrated that intra-tumoral administration of THC to two patients with glioblastoma was decreased expression of VEGF level and its receptor in tumors [47].

Inhibition of tumor metastasis using cannabinoids

Glial cell invasion is a critical event in cancer cell spreading, metastasis and growth of new tumor into other region of brain. Metastasis is a reason for tumor recurrence and death of patient with glioblastoma. In the different studies have reported that cannabinoids decrease adhesion, migration and invasion through various pathways. The signaling pathways are involved in the antimetastatic action of cannabinods including FAK, ERK1/2 and Akt. In signaling pathways of antimetastatic was observed dephosphorylation of ERK1/2 and Akt and phosphorylation of FAK after treatment of cancer cells with THC [51,52]. Tissue inhibitors of alloproteinase-1(TIMP-1) expression were enhanced in malignant glioma. It play important role in tumor metastasis and invasiveness. There is evidence that THC can inhibit TIMP-1 expression in primary tumor cell derived from glioblastoma patients [53]. Together, cannabionids might decrease the migration of tumor cells through signaling pathways and reduction of the TIMP-1 expression.

Clinical approaches and future therapeutic purposes for glioblastoma

Hopeful results obtained from cannabinoids exposed gliomas in *in vitro* and *in vivo* studies. The first clinical trial was performed by the Guzman's research group in 2002 for investigating the antitumoral effects of THC. This group has administrated THC intratumorally to nine patients suffering recurrent glioblastoma for 15 days. Assessment of magnetic resonance imaging indicated that THC delivery reduced tumor growth rate and also psychoactive effects were not observed in these patients [54]. The immunostaining results indicated that THC administration declined proliferation of tumor cell (assessment of Ki67 marker) [54] and enhanced stimulation of autophagy and apoptosis induction in tumor cells (assessment of caspase 3) [36,55]. So, survival median of patients was enhanced for 24 week to 1 year in respect

to reports. These findings were hopeful and it leads to an increase in the interest rate for use of cannabinoids in glioblastoma treatment. However, they claimed that need for more survey in order to cannabinoid use with other anticancer drugs and the suitable selection for administration routes of drug [54]. Researchers have introduced new strategy for glioblastoma treatment such as combination use of cannabinoids with chemotherapeutic drugs [56]. They mentioned that combined treatment of cannabinoid with a classical treatment such as chemotherapy seems to mediate successful results. Study results of Carracedo et al. confirmed this strategy and it can produce the potent and synergic effects via enhanced autophagy (programmed cell death) in brain tumor than either cannabinoids or temozolomide alone [36]. Advantages of the combinational therapies application are production of synchronized effects aiming for inhibition of growth and metastasis of glioma cells. Other hypothesis is combination use of two cannabinoid such as THC with cannabinol for glioblastoma treatment [58]. This way is more effective in the inhibition of tumor growth than THC alone [55,56]. Moreover, simultaneous use of THC and cannabinol together with chemotherapy –temozolomide- could be considered as a therapeutic approach for glioblastoma [56].

Cannabinoids side effects in patients with glioblastoma

According to different studies about cannabinoids have been observed this drug has safety profile. Cannabinids administration is limited in medicine because of their psychoactive effects. Although, it mentioned that psychoactive and adverse effects of cannabinoids might be mediated through CB1 receptor within the brain. Whereas, expression of CB2 receptor was increased in glioblastoma according to cannabinoids target CB2 receptor for inducing apoptosis of glioma cells. Studies have demonstrated that selective CB2 receptor activation declines tumor volume in mice without signs of psychoactive effects [59]. At result, use of selective CB2 receptor agonists such as cannabinol could have contributed to the control of the glioblastoma [60].

CONCLUSION

Together, cannabinoids have been indicated that induce tumor cells apoptosis and inhibit proliferation, angiogenesis and metastasis through activation of its receptors in glioblastoma with the very low toxicity than chemotherapeutic agents. Also, cannabinoids could be applied as single drug or together with anti-cancer

drugs for tumor growth inhibition. Based on these results, cannabinoids could be introduced as acceptable compounds for the control of glioblastoma.

Conflict of Interest

The authors declare no conflict of interest.

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